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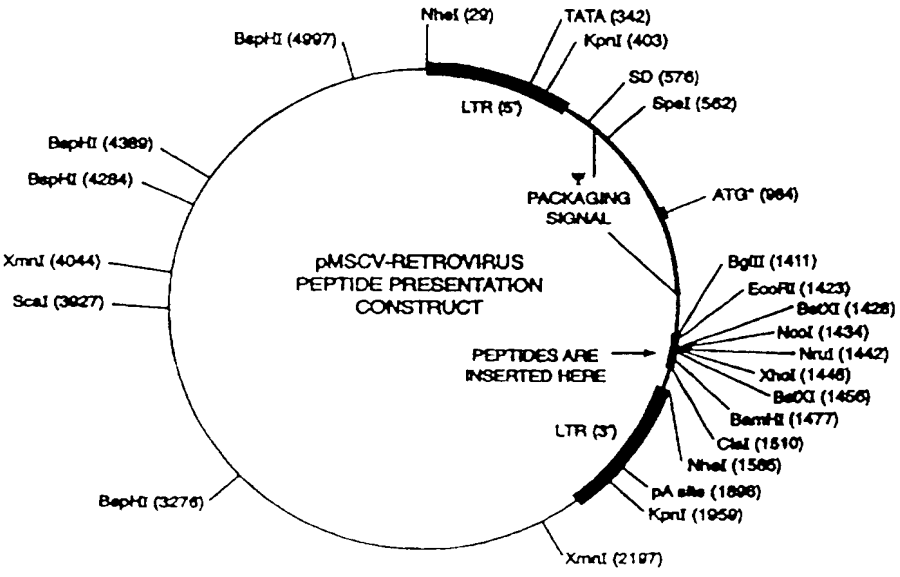
(54) Title: METHODS FOR SCREENING FOR TRANSDOMINANT EFFECTOR PEPTIDES AND RNA MOLECULES



(57) Abstract

Methods and compositions for screening for transdominant effector peptides and RNA molecules selected inside living cells fro randomized pools are provided.

CLAIMS

We claim:

1. A method for screening for a transdominant bioactive agent capable of altering the phenotype of a cell, said method comprising the steps:

a) introducing a molecular library of randomized candidate nucleic acids into a plurality of cells, wherein each of said nucleic acids comprises a different nucleotide sequence;

b) screening said plurality of cells for a cell exhibiting an altered phenotype, wherein said altered phenotype is due to the presence of a transdominant bioactive agent.

2. A method according to claim 1 further comprising the step:

c) isolating said cell exhibiting an altered phenotype.

3. A method according to claim 2 further comprising the step:

d) isolating a candidate nucleic acid from said cell.

4. A method according to claim 2 or 3 further comprising the step:

e) isolating a target molecule using

i) a candidate nucleic acid; or

ii) the expression product of a candidate nucleic acid.

5. A method according to claim 1 wherein said randomized candidate nucleic acids are expressed in said cells to produce a plurality of randomized candidate expression products.

6. A method according to claim 5 wherein said randomized candidate expression products are peptides.

7. A method according to claim 5 wherein said randomized candidate expression products are nucleic acid transcripts.

8. A method according to claim 5 wherein said candidate nucleic acids are linked to fusion partners.

9. A method according to claim 8 wherein said fusion partner comprises a presentation sequence capable of presenting said expression product in a conformationally restricted form.

10. A method according to claim 8 wherein said fusion partner comprises a targeting sequence.

11. A method according to claim 10 wherein said targeting sequence is selected from the group consisting of:
a) a localizing signal sequence capable of constitutively localizing said translation product to a predetermined subcellular locale;
b) a membrane-anchoring signal sequence capable of localizing said translation product to a cellular membrane; and
c) a secretory signal sequence capable of effecting the secretion of said translation product.

12. A method according to claim 8 wherein said fusion partner comprises a targeting sequence and a presentation structure.

13. A method according to claim 1 wherein said introducing is with retroviral vectors.

14. A method according to claim 1 wherein said cells are mammalian cells.

15. A method according to claim 1 wherein said library comprises at least 10^4 different nucleic acids.

16. A method according to claim 1 wherein said library comprises at least 10^5 different nucleic acids.

17. A method according to claim 1 wherein said library comprises at least 10^6 different nucleic acids.

18. A method according to claim 1 wherein said library comprises at least 10^7 different nucleic acids.

19. A method according to claim 1 wherein said library comprises at least 10^8 different nucleic acids.

20. A method for screening for a transdominant bioactive agent capable of altering the phenotype of a cell, said method comprising the steps:

a) introducing a molecular library of randomized candidate nucleic acids into a first plurality of cells, wherein each of said nucleic acids comprises a different nucleotide sequence;

b) contacting said first plurality of cells with a second plurality of cells; and

c) screening said second plurality of cells for a cell exhibiting an altered phenotype.

21. A molecular library of retroviruses comprising at least 10^4 different randomized nucleic acids.

22. A molecular library of retroviruses according to claim 21 comprising at least 10^5 different randomized nucleic acids.

23. A molecular library of retroviruses according to claim 21 comprising at least 10^6 different randomized nucleic acids.

24. A molecular library of retroviruses according to claim 21 comprising at least 10^7 different randomized nucleic acids.

25. A molecular library of retroviruses according to claim 21 comprising at least 10^8 different randomized nucleic acids.

26. A cellular library of mammalian cells containing a molecular library of retroviral constructs, said molecular library comprising at least 10^4 different randomized nucleic acids.

27. A cellular library according to claim 26 wherein said constructs are integrated into the cellular genome.